

30. *Trichoblanius guttulatus* (Bosc).

Examination of several classical examples of *Julus pulchellus*, Leach, shows sufficiently conclusively that they are referable to the blind species *Trichoblanius guttulatus* (Bosc), which was established before Leach's name was given.

Although it does not concern our present study very closely, we may add that it follows, of course, that the specific name *pulchellus* is obsolete and cannot be used for the species furnished with ocelli often referred to under that name. For this animal we must in future use the later designation *Nopoiulus venustus* (Meinert).

## REFERENCES.

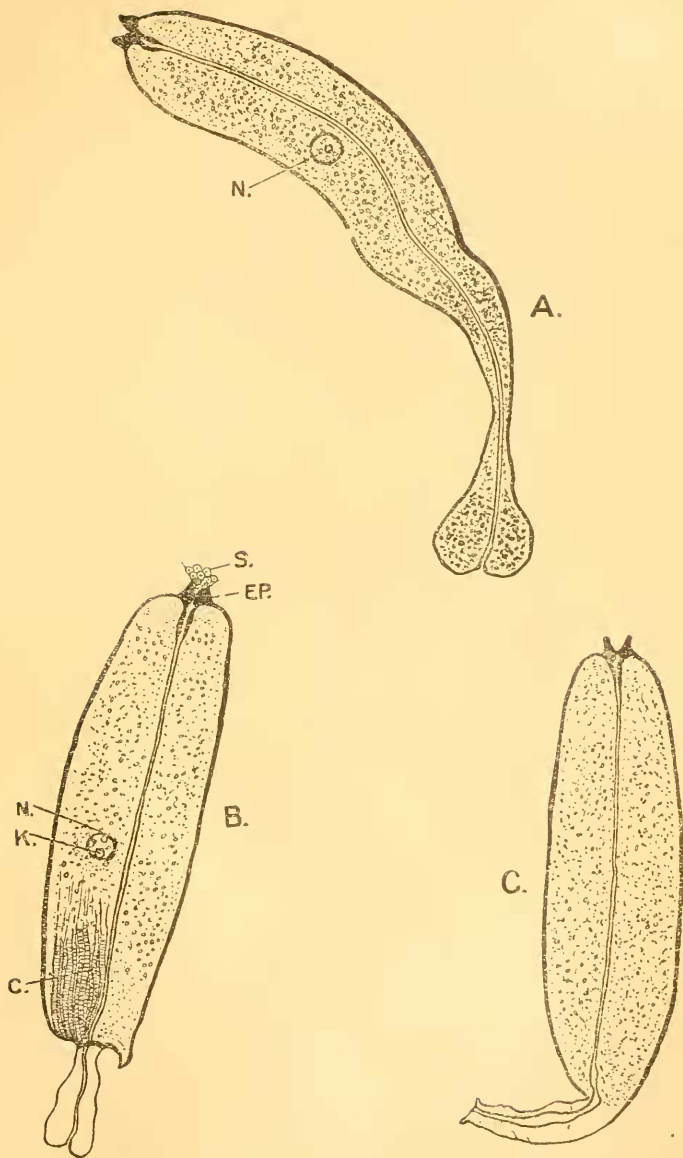
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XXV.—*Some Observations on Pleurocystis cuénoti*, Hesse, 1909, a Cœlozoic Parasite of the Earthworm. By W. HAROLD LEIGH-SHARPE, B.Sc. (Lond.).

ON February 14th, 1918, upon opening a *Helodrilus* (*Allolobophora*) *longus*, Ude, obtained from Red Lion Square, Holborn, London, I discovered seven specimens of diploids (fourteen individuals in all) of the gregarine parasite *Pleurocystis cuénoti*, Hesse, 1909, some of which I have herein figured.

Fig. 1.



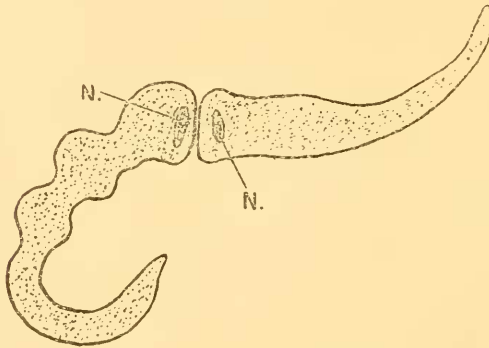
*Pleurocystis cuénoti*. Three diploids, **A, B, C**.

N., nucleus; K., nucleoli; EP., epimerite; C., cytomyomes; S., cells from the seminal funnel of the earthworm.

The parasites reposed in the body-cavity of the worm, in segments 10 and 11, all but one diploid being on the host's right side. They appeared of an opaque china-white colour, the largest (fig. 1, A) being 5 mm. in length, the others 4 mm. All were laterally bowed in shape, and curved round the œsophagus of the worm between the calciferous glands and the pouch, but attached by their anterior extremities to the seminal funnels. The worm had been killed an hour and a half previous to dissection by immersion in methylated alcohol, and none of the parasites showed any sign of life.

It is remarkable that such a large number of parasites should occur within one host, and curious that here we have two individuals permanently associating as one, though not conjugating.

Fig. 2.



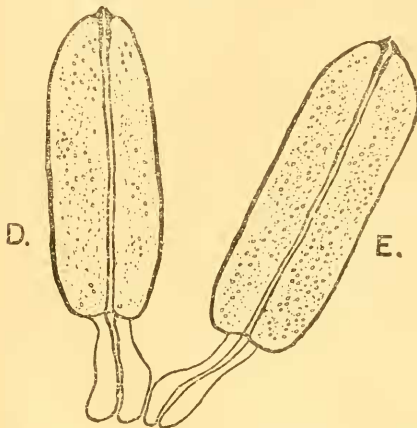
*Monocystis agilis*. Two conjugants in polar apposition.

N., nucleus.

The unique presumed conjugation of *Monocystis magna*, Schmidt, has been described by Cuénot (1900) alone, who gives a figure which, to my mind, is not as satisfactory as might be, since it shows the supposed conjugants so clearly as two separate individuals, instead of being apposed in such a way as to look like one individual (fig. 1), the lines of demarcation between them superficially resembling the alimentary canal of some single animal. The association in this species alone was said to be longitudinal—a fact which appears to have been known at the earliest to Bosanquet (1894), who mentions it casually in describing another species, and even admits *en passant* into his paper a very

inadequate figure. In *Monocystis agilis* and other species the application of the conjugants is polar or "end-to-end" (fig. 2). Since the parasites are attached to the seminal funnels of the hosts permanently, as all agree (and they do not become detached *post mortem*), it is the close proximity of the individuals to one another which determines their association into diploids *inter se*, which must therefore of necessity be longitudinal. A curious confirmation of this is shown in fig. 3 in the case of the diploids D and E, which are partially attached to one another at their posterior extremities, from which I infer that diploidal association probably *always begins at this end*.

Fig. 3.



*Pleurocystis cuénoti*. Two diploids, **D** and **E**, partially attached to one another at their posterior extremities.

But Hesse (1909) pointed out that *Monocystis magna*, Schmidt, differed so materially from other species that he proposed for it the generic name of *Nematocystis*. Further, that there was existing another animal, which, agreeing in the main with the characters of *Monocystis* rather than with those of *Nematocystis*, became coupled, in a longitudinal manner, with a partner for life like *Nematocystis*, and not merely during the process of conjugation. To this form he gave the name of *Pleurocystis cuénoti*, and, while admitting that it is very rare, remarks that the animals are always in the diploidal condition and *never found singly*. After giving

careful consideration to the matter of the claims of *Nematocystis* and *Pleurocystis* as independent genera, I have come to the conclusion that the characters of my specimens agree in the main with those diagnostic of *Pleurocystis* except in regard to a small discrepancy as to size. Hesse states that *Pleurocystis* is 2 mm. long and *Nematocystis* 5 mm., whereas my specimens are of the length of 4-5 mm. If the animal is as rare as Hesse implies, it is possible that it may attain dimensions greater than that mentioned by him, or, again, that an English variety may be larger than the French one which he found in and around Dauphiné. There exists, as far as I can ascertain, no previous record of *Pleurocystis* occurring in the British Isles, though it has possibly been found and confused with the presumed conjugation stage of *Monocystis magna*, which Hesse has now determined is not a conjugation but a life-association into diploids, thereby deciding him to alter its name to *Nematocystis magna*.

The statement of Ceconi (1903) that in *Monocystis agilis*, at any rate, each conjugant separately forms a cyst around itself, which afterwards coalesces with that of its partner, was scouted by Cuénot. Such is not true at any rate for *Pleurocystis*, where the double (cuticular) separation between the individuals is *never broken down*. The cuticle exhibits a network of fine striations, and as the cytoplasm shrinks away from it by plasmolysis, it may perhaps have been mistaken for a cyst. Some of the specimens were ruptured, and others easily became so at the slightest touch. The endoplasm was plentifully stored with paramylum (or paraglycogen). Specimen A, which is also the largest, being 1 mm. longer than any other diploid, is markedly different from the rest. It exhibits a kind of caudal formation, as though it had been killed while making Euglenoid movements; further, the cytoplasmic contents extend right to the posterior extremity, and are more densely granular. Conversely, the other specimens present a normal specific outline, the cytoplasm is withdrawn from the posterior extremity as though shrinkage was occurring previous to cyst-secretion. From these observations I infer that A is an earlier, and possibly the earliest, stage of diploid association, and that the other figures represent subsequent stages. The figures of no other observer represent any such caudal formation.

Though I have figured a nucleus in two individuals, these were not apparent through the cytoplasm; but, upon rupture of the cuticle, when the contents become extruded the nucleus is plainly visible. The nucleus is situated about halfway

along the animal, or anterior to that position (specific character), not at the extreme anterior end as in *Monocystis agilis*, is enclosed in a well-marked nuclear membrane, and contains numerous nucleoli (generic and specific character).

The posterior extremity of B exhibits, under a high magnification, in the inner portion of the ectoplasm, a layer of contractile fibrillæ which have been called myocytes, which I interpret to mean "muscle-cells." As the organism is unicellular, I propose that they be called *cytomyomes*, which I interpret as "cell-muscles."

As this species is a permanently fixed parasite upon the seminal funnels, one might reasonably expect to find some organ of attachment. As to whether one exists there appears to be some difference of opinion, none being mentioned by most of the earlier authors cited for *Nematocystis* (*Monocystis*) *magna*. I am of the definite opinion, however, that the cuticle of the anterior end is prolonged into a blunt petal-like projection (the equivalent of the epimerite of other Gregarines), which stains with micro-carminé like the remainder of the cuticle, is unprovided with hooks, and to which the cells of the seminal funnels of the earthworm are in several cases still adhering. This accords with a passage which Hesse quotes from Bütschli (1882), and not with his own observations, for while he appears to me to describe some organ of attachment, yet he denies that it is in any way an epimerite. I agree with Hesse that it comes in contact, by boring, with many host-cells, and is not attached to one only as he states is the case in *Nematocystis*.

No spermatozoa of the worm were found in the seminal vesicles, or round the seminal funnels, or attached to any of the diploids, such as other observers have figured, nor were any "tails" to be discovered. This may be because the worm had recently shed all its spermatozoa in the regular breeding-season, which obtains about this date, since the seminal vesicles were very small, or because the large number of parasites present had disintegrated and digested them all.

In specimens stained with Ranvier's micro-carminé 25 min. only the cuticle absorbed the carminé, while all the cell-contents became yellow from the action of the picric acid. In specimens stained with Meyer's hæmalum 5 min. both cuticle, cytoplasm, and nucleus became purple, as also the seminal funnel-cells of the host.

The partners of a diploid appear to adhere to one another by a mucilaginous extra-cuticular secretion of "cement"-substance analogous with that which binds the individual

cells of a filament of the alga *Spirogyra* to one another, and is affected by similar stains (e. g. Bismarck brown). It is unaffected by dilute (1 %) acids (cf. the cysts), but the individuals can be made to part from one another by pressure on the coverslip, though not without rupture of the cuticle—which is evidence against their permanent fusion into one organic individual.

#### SUMMARY.

- (1) *Pleurocystis* has established claims to be considered an independent genus.
- (2) It occurs in the same host (*Helodrilus longus*) both in England and France.
- (3) The individuals are in lifelong diploidal association, and not merely at the period of conjugation, as was supposed in *Monocystis magna*, since the nuclei show no signs of division, and it is improbable that seven presumed conjugants should be in the same initial stage.
- (4) The diploids show a caudal formation, but whether this is due to Euglenoid movements or oncoming cyst secretion, or is an artefact, is uncertain.
- (5) I have attempted to set forth reasons for believing that diploidal association commences at the posterior extremity of the individuals.
- (6) The animals attain a greater magnitude than Hesse asserted.
- (7) There is an anterior organ of attachment.
- (8) The partners are firmly but not inseparably attached by an adhesive secretion, and are not permanently fused into one individual.

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